

FAES BioTech 57  
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BLAST, A Basic User's Guide (Wayne Matten)  
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- Intro slides
- Example web BLAST search with 24-mer (TCTAAGTGAAAATCATGACATTTG)
  - Say you have a primer or oligo sequence and you want to...
  - 1) check it's specificity in a genome
  - 2) visualize where it binds
  - Example BLAST: RID = KTY9C75F013
  - Results for KTY9C75F013
    - one full-length hit; answer to #1 - yes, it is unique in human.
    - use 'Graphics' link to view in Sequence viewer; aligns to an exon in ATM gene
- Review BLAST home page and submitting a search:
  - program choice
  - query syntax and number
  - database choice
  - search settings
  - word size (slides)
  - Expect value (slides)
  - Max target sequences
  - "Your search parameters were adjusted to search for a short input sequence."
- Primer-BLAST
  - back to results for KTY9C75F013
    - view SNPs in this region; Tracks -> Variation -> checkboxes, ClinVar
    - primers from selection in Seq viewer
      - Can use to amplify the exon and known SNPs in exon.
      - (Take a wide range in this case; ~ 108302652:108303226)
- Homology search
  - Begin with unknown transcript (TSAseq.txt)
    - to identify, could run blastn vs (nt? refseq\_rna? TSA? 16S? SRA?); organism limit
    - another option: use ORF Finder, then run ORF in SmartBLAST or BLASTP
      - [www.ncbi.nlm.nih.gov/orffinder](http://www.ncbi.nlm.nih.gov/orffinder)
      - For BLASTP, choose database based on goals
    - to find distant relatives, use blastx vs (nr? refseq\_protein?); max target seqs=1000; Expect threshold=1e-6; KU66BD1S013
      - use taxonomy report to explore depth of relatedness
- Resources
  - YouTube Videos: [www.youtube.com/ncbinlm](http://www.youtube.com/ncbinlm)
    - [www.youtube.com/watch?v=KLBE0AuH-Sk](http://www.youtube.com/watch?v=KLBE0AuH-Sk) -> Webinar: A Practical Guide to NCBI BLAST
    - [blast-help@ncbi.nlm.nih.gov](mailto:blast-help@ncbi.nlm.nih.gov)
- Alternatives to web BLAST
  - Standalone; URL API; BLAST in the cloud (see homepage).